

## *Monitored Nondestructively for the First Time by Infrared Spectromicroscopy*

**H**exavalent chromium ( $\text{Cr}^{6+}$ ) is a widespread contaminant that enters the environment through various industrial processes.  $\text{Cr}^{6+}$  compounds are highly water soluble, toxic in the environment, and known carcinogens in mammals. However, recent studies have shown that certain bacterial species can detoxify the compounds by reducing them to relatively insoluble and hence significantly less harmful trivalent chromium ( $\text{Cr}^{3+}$ ) compounds, a process known as biogeochemical transformation. At the Advanced Light Source (ALS), Berkeley Lab scientists Hoi-Ying Holman, Dale Perry, and their colleagues have used the new Fourier-transform infrared (FTIR) spectromicroscopy beamline (Beamline 1.4.3) to obtain spatially resolved, time-dependent evidence for biogeochemical transformation of  $\text{Cr}^{6+}$  on magnetite and Columbia River basalt. This is the first time that biogeochemical transformation of  $\text{Cr}^{6+}$  on a mineral surface has been nondestructively monitored and studied.

Although infrared spectra are not historically associated with synchrotrons, the researchers point out that

the relatively new synchrotron-radiation FTIR technique (SR-FTIR) is perfect for monitoring biogeochemical processes. Because infrared does not kill bacteria, the transformation of  $\text{Cr}^{6+}$  can be mapped as it occurs. Distinct and relevant infrared absorption bands related to the transformation are used not only as chemical markers to detect different chromium species, but also as biological markers to detect the presence and activity of microorganisms on specimen surfaces. In addition, the brightness of the infrared radiation makes spatially resolved spectroscopy (spectromicroscopy) possible.

Two reduction mechanisms had previously been postulated for the reduction of hexavalent chromium compounds. The biological mechanism requires the presence of microorganisms to aerobically reduce the  $\text{Cr}^{6+}$ . The chemical mechanism relies on metal oxides, such as Fe(II) compounds, to catalyze the  $\text{Cr}^{6+}$ -reduction reaction. The Berkeley Lab researchers conducted two SR-FTIR experiments to distinguish the relative significance of these two mechanisms. In addition, they evaluated effects of

common organic co-contaminants, such as toluene vapor, on the biotic reduction process.

The first experiment examined the biotic and abiotic mechanisms on magnetite surfaces. After a five-day exposure to  $\text{Cr}^{6+}$  compounds on samples containing no living microorganisms, the absence of statistically significant changes in infrared chemical and biological markers indicated that no catalysis of  $\text{Cr}^{6+}$  reduction was occurring. On samples with living microorganisms, however, some  $\text{Cr}^{6+}$  reduction was detected. Moreover, when the samples with living microorganisms were incubated in toluene vapor, statistically significant changes in both infrared-absorption intensity and characteristic band shapes were observed for  $\text{Cr}^{6+}$ , as were new bands signaling the possible existence of intermediate  $\text{Cr}^{5+}$ , all indicating catalysis of substantial  $\text{Cr}^{6+}$  reduction. With SR-FTIR spectromicroscopy, the researchers showed that the changes in the infrared absorption bands occurred at the sites of bacterial concentration. Imaging the surface at characteristic absorption bands showed a strong correlation between

peak depletion of  $\text{Cr}^{6+}$  and toluene and peak concentration of biological molecules.

The second experiment was a four-month study to determine if this microbial reduction process could occur in real geologic samples. Composite mineral surfaces of basalt rock chips containing resident communities of microbes were exposed to solutions of hexavalent chromium and toluene vapor. At the end of four months, SR-FTIR spectromicroscopy showed that  $\text{Cr}^{6+}$ -tolerant and  $\text{Cr}^{6+}$ -reducing natural microorganisms were thriving in association with  $\text{Cr}^{3+}$ . The reduced  $\text{Cr}^{3+}$  state was confirmed by x-ray absorption fine structure (XAFS) spectroscopy. These  $\text{Cr}^{3+}$  compounds remained stable at the end of the experiment.

These nondestructive infrared spectromicroscopy studies, combined with microbiological techniques, show that highly mobile and toxic  $\text{Cr}^{6+}$  contaminants can be biologically reduced into less soluble, less toxic  $\text{Cr}^{3+}$  compounds. The SR-FTIR method can now be expanded to examine other infrared-amenable microbial/chemical contaminant systems.

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H.-Y. N. Holman, D. L. Perry, M. C. Martin, G. M. Lamble, W. R. McKinney, and J. C. Hunter-Cevera. "Real-Time Characterization of Biogeochemical Reduction of Cr(VI) on Basalt Surfaces by SR-FTIR Imaging," *Geomicrobiology J.*, **16**(4) (1999) in press.

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# MICROBIAL REDUCTION OF HEXAVALENT CHROMIUM

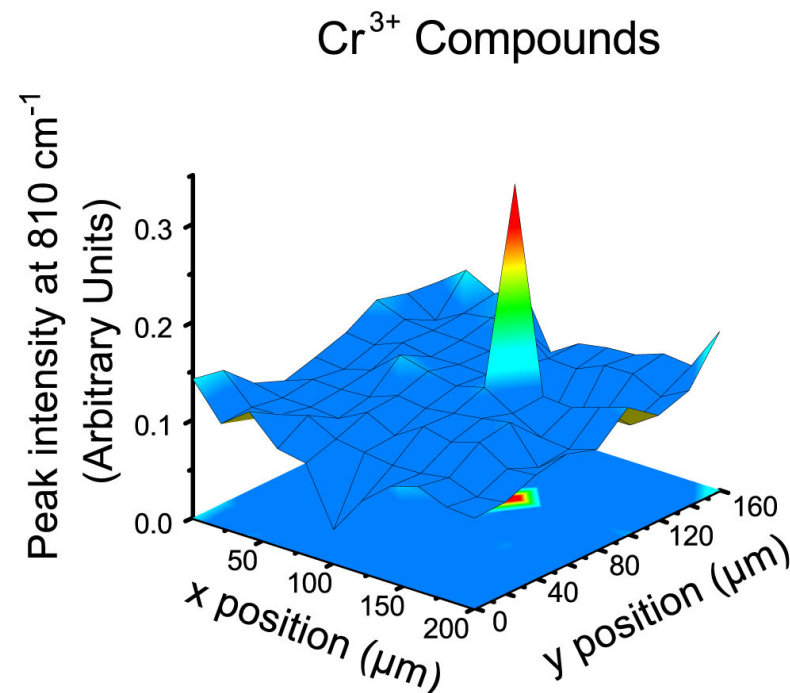
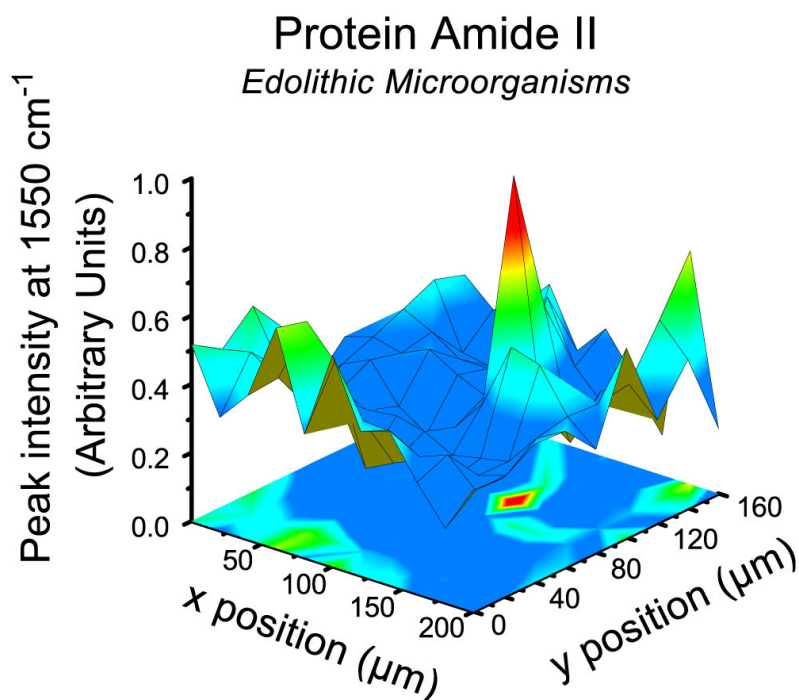


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- **Hexavalent chromium ( $\text{Cr}^{6+}$ ) compounds in the environment**
  - *Widespread, highly mobile, mutagenic contaminants from industrial waste*
  - *Reduction to trivalent  $\text{Cr}^{3+}$  results in immobile, less harmful compounds*
- **Two mechanisms for reduction — which is dominant?**
  - *Reduction by microorganisms (biogeochemical transformation)*
  - *Catalysis by metal-oxides or natural organic molecules (chemical reduction)*
- **Synchrotron-Radiation FTIR spectromicroscopy is ideal tool**
  - *Real-time monitoring possible with live microorganisms*
  - *Distinctive absorption bands act as chemical, biological markers*
  - *Spatial resolution localizes transformation process*
- **Berkeley Lab experiments demonstrate biogeochemical reduction**
  - *Biological reduction dominates in both lab and natural samples*
  - *Sites of  $\text{Cr}^{6+}$  and toluene depletion linked to location of microorganisms*
  - *Sites of  $\text{Cr}^{3+}$  concentration associated with naturally occurring bacteria*

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